Reviewed by: Robert F. Fricke, Ph.D. Robert F. Fricke 21 Dec 94
Section II, Tox. Branch II (7509C)

Secondary Review: Clark Swentzel K. Clark Swentzel 12/21/94
Section II, Tox. Branch II (7509C)

DATA EVALUATION REPORT

STUDY TYPE: Acute Oral Toxicity - Rats (§81-1)

EPA ID NOs.: MRID No.: 434410-37
PC Code: 129122
DP Barcode: D209640
Submission No.: S477407

TEST MATERIAL: BF-309

SYNONYMS: XDE-564, N-(2,6-dichlorophenyl)-5-ethoxy-7-fluoro[1,2,4]triazolo-[1,5-c] pyrimidine-2-sulfonamide

STUDY ID NO.: DR-0342-6720-001A

SPONSOR: DowElanco
9330 Zionville Road
Indianapolis, IN

TESTING LAB: The Toxicology Research Laboratory
Health and Environmental Sciences
The Dow Chemical Company, Midland, MI

REPORT TITLE: BF-309: Acute Oral Toxicity Study in Fischer 344 Rats

AUTHOR: K.S. Gilbert

REPORT ISSUED: 3 October 1994

EXECUTIVE SUMMARY: A single group of Fischer 344 rats (5/sex) was orally gavaged with a 50% aqueous suspension of BF-309 (84.3% XDE-564, a.i.) at a single dose of 5000 mg/kg. All of the animals survived until terminal sacrifice. Fecal and/or urine soiling were seen through Day 4 in females and Day 5 in males; salivation was observed through Day 2 in males and Day 3 in females. All animals gained body weight during the study. No treatment-related changes were observed at gross pathological examination.

LD_{50} > 5000 mg/kg (limit dose) for males and females

Toxicity category IV

This study is classified as **Core - Guideline** and satisfies guideline requirements (§81-1) for an acute oral toxicity study in the rat.

MATERIALS AND METHODS

Test Compound: BF-309 Description: Solid Reference No.: TSN100373 Purity: 84.3% a.i. (XDE-564) Contaminants: Not given.

Test Animals: Species: Rat Strain: Fischer 344 Age: 8 weeks Weight (g): 189.5 - 206.4 (males), 129.3 - 136.2 (females) Source: Charles River Breeding Laboratories, Inc., Kingston, NY Food: Purina Certified Rodent Chow # 5002, *ad libitum* Water: Tap water, *ad libitum* Environmental: Temperature: 22 ± 3, Relative humidity: 40 - 70%, Air changes: 15/hr, Light/dark photocycle: 12hr/12hr.

Study Design: A single group of Fischer 344 rats (5/sex) were orally gavaged with a 50% aqueous suspension of BF-309 (84.3% a.i., XDE-564) at a single dose of 5000 mg/kg. Animals were observed for 15 days for clinical signs of toxicity, mortality and moribundity. Animals were observed for signs of toxicity, moribundity and mortality immediately after dosing (1, 4, 5½, and 7½ hr) and on Days 2, 3, 4, 5, 8, 9, 10, 11, 12, and 15. Animals were weighed at prestudy and on Days 2, 8, and 15 of the observation period. At the end of the observation period animals were necropsied for gross pathological examination.

Statistics: Body weight data were expressed as means and standard deviations. Grubbs method was used to evaluate data for outliers.

RESULTS AND CONCLUSIONS

A single group of Fischer 344 rats (5/sex) were orally gavaged with a 50% aqueous suspension of BF-309 (84.3% a.i., XDE-564) at a single dose of 5000 mg/kg. All of the animals survived until terminal sacrifice. Fecal and/or urine soiling were observed through Day 4 in females and Day 5 in males; salivation was observed through Day 2 in males and Day 3 in females. All animals gained body weight during the study. No treatment-related changes were observed at gross pathological examination.

LD_{50} > 5000 mg/kg (limit dose) for males and females

Toxicity Category IV

Classification: core - Guideline

This study satisfies guideline requirements (§81-1) for an acute oral toxicity study in the rat.

Reviewed by: Robert F. Fricke, Ph.D. Robert F. Fricke 21 Dec 94
Section II, Tox. Branch II (7509C)

Secondary Review: Clark Swentzel R. Clark Swentzel 12/21/94
Section II, Tox. Branch II (7509C)

DATA EVALUATION REPORT

STUDY TYPE: Acute Dermal Toxicity - NZW Rabbits (§81-2)

EPA ID NOs.: MRID No.: 434410-38
PC Code: 129122
DP Barcode: D209640
Submission No.: S477407

TEST MATERIAL: BF-309

SYNONYMS: XDE-564, N-(2,6-dichlorophenyl)-5-ethoxy-7-fluoro-[1,2,4]triazolo-[1,5-C] pyrimidine-2-sulfonamide

STUDY NO.: DR-0342-6720-001D

SPONSOR: DowElanco
9330 Zionville Road
Indianapolis, IN

TESTING LAB: The Toxicology Research Laboratory
Health and Environmental Sciences
The Dow Chemical Company, Midland, MI

REPORT TITLE: BF-309: Acute Dermal Toxicity Study in New Zealand White Rabbits

AUTHOR: K.S. Gilbert

REPORT ISSUED: 3 October 1994

EXECUTIVE SUMMARY: A single group of NZW rabbits (5/sex) was dermally exposed for 24 hr to BF-309 at a dose of 2000 mg/kg. All animals survived to terminal sacrifice. Except for the presence of erythema in all animals at the end of the dosing period, no other clinical signs of toxicity were observed. All animals gained body weight during the study. No treatment-related gross pathological findings were observed.

LD_{50} > 2000 mg/kg (limit dose) for males and females

Toxicity Category III

This study is classified as **Core - Guideline** and satisfies guideline requirements (§81-2) for an acute dermal toxicity study in the rabbit.

MATERIALS AND METHODS

Test Compound: BF-307 Description: Solid Reference No.: TSN100373 Purity: 84.3% a.i. (XDE-564) Contaminants: Not given.

Test Animals: Species: Rabbit Strain: New Zealand White Age: 4 months Weight (kg): 2.20 - 2.60 (males), 2.42 - 2.58 (females) Source: Hazelton Research Products, Kalamazoo, MI Food: 4 oz/day of Purina Certified Rabbit Chow #5322, *ad libitum* Water: Tap water, *ad libitum* Environmental: Temperature: 19 ± 3°C, Relative Humidity: 40 - 60%, Light/dark cycle: 12 hr/12 hr, Air changes: 15/hr.

Study Design: A single group of NZW rabbits (5/sex) was dermally exposed to BF-309 at a dose of 2000 mg/kg. BF-309 was moistened with 2 ml of distilled water and applied to a piece of gauze (10 x 14 cm, 10% of body surface area). The gauze was placed on a fur-free area of the back and held in place with an elastic rabbit jacket. After 24 hr, the jacket and gauze were removed and the application site thoroughly cleaned with water and dried. Animals were observed for signs of toxicity during dosing and daily, thereafter, for 15 days. Animals were weighed during prestudy and on Days 2, 8, and 15. At the end of the observation period animals were necropsied for gross pathological examination.

Statistics: Body weight data were expressed as means and standard deviations. Grubbs method was used to evaluate data for outliers.

RESULTS AND CONCLUSIONS

A single group of NZW rabbits (5/sex) were dermally exposed to BF-309 at a dose of 2000 mg/kg for 24 hr. All animals survived to terminal sacrifice. Except for the presence of erythema in all animals at the end of the dosing period, no other clinical signs of toxicity were observed. All animals gained body weight during the study. No remarkable changes related to the test material were observed in gross pathological examination.

LD_{50} > 2000 mg/kg (limit dose) for males and females

Toxicity Category III

Classification: core - Guideline

This study satisfies guideline requirements (§81-1) for an acute oral toxicity study in the rat.

011435

Reviewed by: Robert F. Fricke, Ph.D. Robert F. Fricke 21 Dec 94
Section II, Tox. Branch II (7509C)

Secondary Review: Clark Swentzel R. Clark Swentzel 12/21/94
Section II, Tox. Branch II (7509C)

DATA EVALUATION REPORT

STUDY TYPE: Acute Inhalation Toxicity - Rats (§81-3)

EPA ID NOs.: MRID No.: 434410-39
PC Code: 129122
DP Barcode: D209640
Submission No.: S477407

TEST MATERIAL: BF-309

SYNONYMS: XDE-564, a.i.
[1,2,4] Triazolo-[1,5-c]pyrimidine-2-sulfonamide: N-(2,6-dichlorophenyl)-5-ethoxy-7-fluoro-

STUDY ID NO.: DR-0342-6720-002

SPONSOR: DowElanco
9330 Zionville Road
Indianapolis, IN

TESTING LAB: The Toxicology Research Laboratory
Health and Environmental Sciences
The Dow Chemical Company, Midland, MI

REPORT TITLE: BF-309: Acute Aerosol Inhalation Study with Fischer 344 Rats

AUTHOR: F.S. Cieszlak and C.M. Clements

REPORT ISSUED: 30 September 1994

EXECUTIVE SUMMARY: A single group of Fischer 344 rats (5/sex) was exposed to a dust aerosol of BF-309 at a concentration of 6.70 mg/l. The particle size distribution of the aerosol had an MMAD ± σ_g of 3.63 ± 2.29 μm. The total mass of particles having diameters less than 1 μm was approximately 8%, and less than 6 μm, 74%. All animals survived to terminal sacrifice without any treatment-related clinical signs or changes in mean body weights. Necropsy on Day 15 did not show any gross pathological findings which could be attributed to treatment.

LC_{50} > 6.7 mg/l (limit dose) for males and females

Toxicity category IV

This study is classified as **Core - Guideline** and satisfies guideline requirements (§81-3) for an acute aerosol toxicity study in the rat.

MATERIALS AND METHODS

Test Compound: BF-309 Description: Off-white powder Lot Nos.: A-840-91, TSN100373 Purity: 84.3% (86% XDE-564 at 97% purity) Contaminants: Not given.

Test Animals: Species: Rat Strain: Fischer 344 Age: 13 weeks Weight (g): 238.6 - 269.9 (males), 141.2 - 165.1 females) Source: Charles River Breeding Laboratories, Inc., Kingston, NY Food: Purina Certified Rodent Chow #5002 Water: Tap water, *ad libitum* Environmental: Temperature: 22 ± 3°C, Relative humidity: 40 - 70%, Light/dark photocycle: 12 hr/12 hr, Air changes: 15/hr.

Study Design: A single group of Fischer 344 rats (5/sex) was exposed, using a nose-only chamber, to a powder aerosol of BF-309 at a concentration of 6.70 mg/l for four hours. Animals were acclimated to the nose-only chamber during a four-hour period before the initiation of the study. Animals were observed during the exposure and daily, thereafter, for 15 days. Animals were weighed at prestudy and on Days 2, 4, 8, 11, and 15. All animals were necropsied for gross pathological examinations.

Test Atmosphere Generation and Test Chamber Conditions: Dust aerosols were generated from powdered BF-309, which was uniformly fed into an air mill using an auger screw. The milled test material was passed through a cyclone and into a 42 liter, nose-only exposure chamber. Temperature, humidity, and air flow were measured at the beginning of the exposure and at 30-minute intervals, thereafter, for 4 hr; gravimetric analysis of the chamber atmosphere was performed at 45, 130, and 200 min of the exposure period.

Statistics: Body weight data were expressed as means and standard deviations.

RESULTS

Test Chamber Conditions and Aerosol Characterization: The mean (± S.D.) temperature and relative humidity within the exposure chamber were 20.2 ± 0.3°C (n = 9) and 61.8 ± 0.4% (n = 9), respectively. Air flow through the chamber was maintained at 30 l/min. The time weighted concentration in the chamber was 6.70 mg/l. Particle size data for this study are summarized in Table 1.

Clinical Observations and Mortality: All animals survived to terminal sacrifice without the appearance of treatment-related clinical signs.

Mean Body Weights: Mean body weights were slightly reduced (2.4%, males; 1.1%, females) on study Day 2.

Gross Pathology: Necropsy, performed on Day 15, did not reveal any abnormal gross pathological changes, the incidences of findings were within normal limits.

Table 1: Particle size distribution data (Summarized from Table 2 of the study)

Effective Cutoff Diameter (μm)	Cumulative % Less than Stated Size
0.68	5.79
1.3	9.38
3	23.93
6.1	74.07
11	92.98
17	96.61
MMAD $\pm$ σ_g	3.63 $\pm$ 2.29 μm

CONCLUSIONS

Animals (5 rats/sex) were exposed for four hours in a nose-only chamber to a dust aerosol of BF-309 at a concentration of 6.70 mg/l. The particle size distribution of the aerosol had an MMAD $\pm$ σ_g of 3.63 $\pm$ 2.29 μm. The total mass of particles having diameters less than 1 μm was approximately 8%, and less than 6 μm, 74%. All animals survived to terminal sacrifice without any treatment-related clinical signs or changes in mean body weights. Necropsy on Day 15 did not show any gross pathological findings which could be attributed to treatment.

LC_{50} > 6.70 mg/l

Toxicity category IV

Classification: core - Guideline

This study satisfies guideline requirements (§81-3) for an acute inhalation toxicity study in the rat.

011433

Reviewed by: Robert F. Fricke, Ph.D. Robert F. Fricke 21 Dec 94
Section II, Tox. Branch II (7509C)

Secondary Review: Clark Swentzel K. Clark Swentzel 12/22/94
Section II, Tox. Branch II (7509C)

DATA EVALUATION REPORT

STUDY TYPE: Primary Eye Irritation - NZW Rabbits (§81-4)

EPA ID NOs.: MRID No.: 434410-40
PC Code: 129122
DP Barcode: D209640
Submission No.: S477407

TEST MATERIAL: BF-309

SYNONYMS: XDE-564
N-(2,6-dichlorophenyl)-5-ethoxy-7-fluoro-[1,2,4] triazolo-[1,5-C] pyrimidine-2-sulfonamide

STUDY NO.: DR-0342-6720-001C

SPONSOR: DowElanco
9330 Zionville Road
Indianapolis, IN

TESTING LAB: The Toxicology Research Laboratory
Health and Environmental Sciences
The Dow Chemical Company, Midland, MI

REPORT TITLE: BF-309: Primary Eye Irritation Study in New Zealand White Rabbits

AUTHOR: K.S. Gilbert

REPORT ISSUED: 3 October 1994

EXECUTIVE SUMMARY: The eye irritation potential of BF-309 was evaluated in NZW rabbits (4 males, 2 females). BF-309 (100 mg) was instilled into the conjunctival sac of the right eye, the left eye served as the control. Ocular irritation was evaluated at 1, 24, 48, and 72 hr and 7 days after treatment. At 24 hr, slight to moderate conjunctival irritation (reddening, 5/6; chemosis, 4/6; discharge, 3/6) and reddening of the iris (3/6) were present; there were no signs of corneal involvement. Signs of ocular irritation persisted through 72 hr post-treatment with complete clearing by Day 7.

Toxicity category III

Classification: core - Guideline

This study satisfies guideline requirements (81-4) for a primary eye irritation study in rabbits.

MATERIALS AND METHODS

Test Compound: BF-309 Description: Solid Reference No.: TSN 100373 Purity: 84.3% Contaminants: Not given.

Test Animals: Species: Rabbit Strain: New Zealand White Age: 17 - 18 months Weight (kg): 2.51 - 2.72 (males), 2.91 - 3.06 (females) Source: Hazelton Research Products, Kalamazoo, MI Food: 4 oz/day of Purina Certified Rabbit Chow #5322 Water: Tap water, *ad libitum* Environmental: Temperature: 19 ± 3°C, Relative humidity: 40 - 60%, Air changes: 15/hr, Light/dark photocycle: 12 hr/12 hr.

Study Design: Four male and two female rabbits were used in this primary eye irritation study. Both eyes were examined before the start of the study, only defect-free rabbits were used. BF-309 (100 mg) was instilled into the conjunctival sac of the right eye, the left eye was untreated and served as the control. Both eyes were examined at 1, 24, 48 and 72 hr and 7 days after treatment for signs of ocular lesions, which were graded and scored using the Draize method. Rabbits were weighed on the day of treatment and at study termination.

RESULTS AND CONCLUSIONS

The ocular reactions to BF-309 are summarized in Table 1. Signs of ocular irritation persisted through 72 hr post-treatment with complete clearing at Day 7. No corneal involvement was present.

Table 1: Ocular Responses to BF-309 (Data summarized from study Table 1)

Response	Incidence of Ocular Responses to BF-309				
	1 hr	24 hr	48 hr	72 hr	7 days
Corneal opacity	0/6 (6,0,0)[a]	0/6 (6,0,0)	0/6 (6,0,0)	0/6 (6,0,0)	0/6 (6,0,0)
Reddening of the iris	0/6 (6,0,0)	3/6 (3,3,0)	1/6 (5,1,0)	1/6 (5,1,0)	0/6 (6,0,0)
Conjunctiva					
Redness	6/6 (0,5,1)	5/6 (1,2,3)	5/6 (1,2,3)	5/6 (1,4,1)	0/6 (6,0,0)
Chemosis	6/6 (0,2,4)	4/6 (2,1,3)	4/6 (2,3,1)	2/6 (4,2,0)	0/6 (6,0,0)
Discharge	0/6 (6,0,0)	3/6 (3,1,2)	1/6 (5,1,0)	0/6 (6,0,0)	0/6 (6,0,0)

[a] Values in parentheses are the incidence of the responses by grade (No response, Grade 1, Grade 2)

Toxicity category III

Classification: core - Guideline

011433

Reviewed by: Robert F. Fricke, Ph.D. Robert F. Fricke 21 Dec 94
Section II, Tox. Branch II (7509C)

Secondary Review: Clark Swentzel K. Clark Swentzel 12/22/94
Section II, Tox. Branch II (7509C)

DATA EVALUATION REPORT

STUDY TYPE: Primary Dermal Irritation (§81-5)

EPA ID NOs.: MRID No.: 434410-41
PC Code: 129122
DP Barcode: D209640
Submission No.: S477407

TEST MATERIAL: BF-309

SYNONYMS: XDE-564, a.i.
N-(2,6-dichlorophenyl)-5-ethoxy-7-fluoro-[1,2,4] triazolo-[1,5-c] pyrimidine-2-sulfonamide

STUDY ID NO.: DR-0342-6720-001B

SPONSOR: DowElanco
9330 Zionville Road
Indianapolis, IN

TESTING LAB: The Toxicology Research Laboratory
Health and Environmental Sciences
The Dow Chemical Company, Midland, MI

REPORT TITLE: BF-309: Primary Dermal Irritation Study in New Zealand White Rabbits

AUTHOR: K.S. Gilbert

REPORT ISSUED: 3 October 1994

EXECUTIVE SUMMARY: The dermal irritation potential of BF-309 was evaluated in NZW rabbits (3/sex). Within 30 min after removal of the test compound, very slight erythema was observed in all of the test animals. At 24 hr, well defined erythema was present in 2/6 animals and very slight erythema in 2/6 animals. Very slight erythema was observed in 3/6 animals at 48 and 72 hr and 2/6 animals at Day 6. By Day 7, erythema was no longer present. Slight edema was observed in only one animal at the 24 hr examination. The primary irritation index at 72 hr equaled 0.75 (slight dermal irritant).

Toxicity category IV

Classification: core - Guideline

This study satisfies guideline requirements (§81-5) for a primary dermal irritation study in rabbits.

MATERIALS AND METHODS

Test Compound: BF-309 Description: Solid Reference No.: TSN100373 Purity: 84.3% a.i. (XDE-564) Contaminants: Not given.

Test Animals: Species: Rabbit Strain: New Zealand White Age: 15-16 months Weight (kg): 2.34 - 2.38 (males), 2.25 - 2.46 (females) Source: Hazelton Research Products, Kalamazoo, MI Food: 4 oz/day of Purina Certified Rabbit Chow #5322 Water: Tap water, *ad libitum* Environmental: Temperature: 19 ± 3°C, Relative humidity: 40 - 60%, Air changes: 15/hr, Light/dark photocycle: 12 hr/12 hr.

Study Design: The dermal irritation potential of BF-309 was evaluated in NZW rabbits (3/sex). The application sites (10 cm x 10cm) on the backs of the animals were clipped free of fur. A 0.5 g aliquot of test compound, moistened with 0.2 ml of water, was applied to a 25 mm Hill Top Chamber and secured in place with an elastic rabbit jacket. The test compound remained in contact with the skin for 4 hours, at which time the jackets and chambers were removed and the application sites cleaned with water to remove any remaining residual test compound. Application sites were then scored for edema and erythema, using the Draize method, within 30 min; 24, 48 and 72 hr; and on Days 6 and 7. Body weights were determined on Days 1 and 7.

RESULTS AND CONCLUSIONS

Within 30 min after removal of the test compound, very slight erythema was observed in all of the test animals. At 24 hr, well defined erythema was present in 2/6 animals and very slight erythema, in 2/6 animals. Very slight erythema was observed in 3/6 animals at 48 and 72 hr and 2/6 animals at Day 6. By Day 7, erythema was no longer present. Slight edema was observed in only one animal at the 24 hr examination. Individual dermal irritation scores are summarized in Table 1. The mean dermal irritation scores and the primary irritation indices (PII) are summarized in Table 2. At the 72 hr, the PII equaled 0.75, indicative of slight dermal irritant.

Toxicity category IV

Classification: core - Guideline

This study satisfies guideline requirements (§81-5) for a primary dermal irritation study in rabbits.

Table 1: Dermal Responses to BF-309 (Data summarized from study Table 1)

Skin Irritation Scores[a]					
≤ 30 min	24 hr	48 hr	72 hr	Day 6	7 days
Erythema					
6/6 (0,6,0)	4/6 (2,2,2)	3/6 (3,3,0)	3/6 (3,3,0)	2/6 (4,2,0)	0/6 (6,0,0)
Edema					
0/6 (6,0,0)	1/6 (5,0,1)	0/6 (6,0,0)	0/6 (6,0,0)	0/6 (6,0,0)	0/6 (6,0,0)

[a] Values in parentheses are the incidence of the responses by grade (No response, Grade 1, Grade 2)

Table 1: Dermal Irritation Results (Summarized from individual dermal irritation grades from study Table 1)

Evaluation Time	Dermal Irritation Scores[a]		Primary Irritation Index[b]
	Erythema	Edema	
< 30 min	1.0	0.0	1.5
24 hr	1.0	0.33	2.0
48 hr	0.5	0.0	0.75
72 hr	0.5	0.0	0.75
6 days	0.33	0.0	0.5
7 days	0.0	0.0	0.0

[a] Mean of individual scores of six rabbits was calculated by the reviewer.
[b] Calculated by reviewer

011433

Reviewed by: Robert F. Fricke, Ph.D. Robert F. Fricke 23 Dec 94
Section II, Tox. Branch II (7509C)

Secondary Review: Clark Swentzel K. Clark Swentzel 1/3/95
Section II, Tox. Branch II (7509C)

DATA EVALUATION REPORT

STUDY TYPE: Dermal Sensitization (§81-6)

EPA ID NOs.: MRID No.: 434410-42
PC Code: 129122
DP Barcode: D209640
Submission No.: S477407

TEST MATERIAL: BF-309

SYNONYMS: XDE-564, a.i.
N-(2,6-dichlorophenyl)-5-ethoxy-7-fluoro-[1,2,4] triazolo-[1,5-c] pyrimidine-2-sulfonamide

STUDY ID NO.: DR-0342-6720-001E

SPONSOR: DowElanco
9330 Zionville Road
Indianapolis, IN

TESTING LAB: The Toxicology Research Laboratory
Health and Environmental Sciences
The Dow Chemical Company, Midland, MI

REPORT TITLE: BF-309: Dermal Sensitization Potential in the Hartley Albino Guinea Pig

AUTHOR: K.S. Gilbert

REPORT ISSUED: 3 October 1994

EXECUTIVE SUMMARY: Buehler's method was used to evaluate the skin sensitivity of the test compound in guinea pigs. Based on the results of the study, BF-309 did not exhibit any skin sensitization potential.

Classification: core - Guideline

This study satisfies guideline requirements (§81-6) for a dermal sensitization study in guinea pigs.

MATERIALS AND METHODS

Test Compound: BF-309 Description: Solid Reference No.: TSN100373 Purity: 84.3% a.i. (XDE-564) Contaminants: Not given.

Test Animals: Species: Guinea pig Strain: Hartley Albino Age: 5-6 months Weight (g): 363 - 444 (males) Source: Charles River Breeding Laboratories, Inc., Kingston, NY Food: Purina Certified Guinea Pig Chow #5026, *ad libitum* Water: Tap water, *ad libitum* Environmental: Temperature: 22 ± 3°C, Relative humidity: 40 - 70%, Air changes: 15/hr, Light/dark photocycle: 12 hr/12 hr.

Preliminary Dermal Irritation Study: A preliminary dermal irritation study was carried out to determine the concentration of BF-309 to be used in the main study. On the day prior to dosing, the backs of animals were clipped free of fur. BF-309 (0.4 g, moistened with 0.2 ml of distilled water) or suspensions (40, 60 or 80% in distilled water) of BF-309 were applied on the backs in 25 mm Hill Top Chambers. Following a 6-hour exposure, the chambers were removed and the application sites cleaned of residual BF-309. Dermal irritation was evaluated 24 and 48 hr post-exposure.

Based on the results of the preliminary study, the highest non-irritating dose was an 80% suspension of BF-309; the highest irritating dose was 0.4 g of 100% BF-309, moistened with 0.2 ml of distilled water.

Main Study

Induction Phase: Animals (10/group) were randomly assigned to treatment and positive control groups. One day prior to dosing, the left side of the back was clipped free of fur. The treatment group received 100% BF-309 ml (0.4 g, moistened with 0.2 ml of distilled water), while the positive control animals, an aliquot of a 10% solution of DER 331 epoxy resin in dipropylene glycol nonomethyl ether (DPGNE). Treatment and positive control materials were applied to the skin using 25 mm Hill Top Chambers, which were secured in place with Vetrap® and covered with Elasticon®. The chambers were removed after a 6-hour exposure and the application sites evaluated for dermal irritation the next day. This same procedure was repeated weekly, for three weeks.

Challenge Phase: Following a two-week, treatment-free period, the right side of the back was clipped free of fur. Treatment group animals were challenged with a 0.4 ml aliquot of an 80% suspension of BF-309 in distilled water, while the positive control group animals, with a 10% solution of DER 331 in DPGME. Five naive control animals each received either the 80% suspension of BF-309 in distilled water or 10% DER 331. Challenge materials were applied using the Hill Top Chambers, as described above.

The chambers were removed after a 6-hour exposure. The application sites were depilated and evaluated for signs of irritation approximately 24 and 48 hr after removal of the chambers.

Scoring: A dermal irritation score of 1.0 or greater at 48 hr in two or more animals was considered a positive response. Naive control animals should show no signs of dermal irritation.

RESULTS AND CONCLUSIONS

In this dermal sensitization study in guinea pigs, BF-309 was evaluated using Buehler's method. No dermal sensitization was noted in any of the treatment or naive control animals. The positive control animals responded appropriately, with slight and moderate responses noted in 5/10 and 4/10 animals, respectively, at 48 hr.

Based on the results of the study, the BF-309 did not exhibit any sensitization potential.

Classification: core - Guideline

This study satisfies guideline requirements (81-6) for a dermal sensitization study in guinea pigs.